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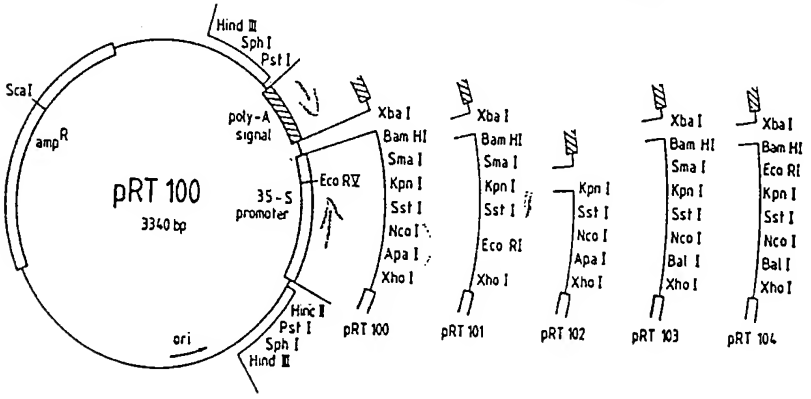
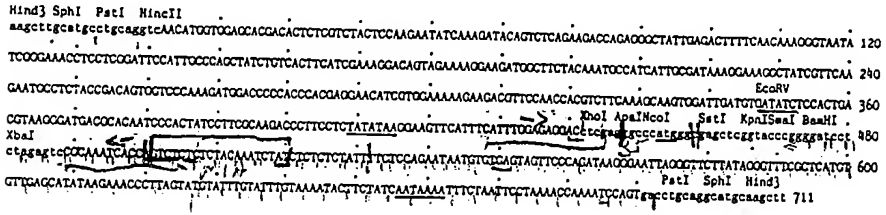
The plasmids pRT100-pRT104, carrying the 35S promoter and the polyadenylation signal of CaMV strain Cabb B-D (corresponding to bp 7016-7434 and 7436-7639 of CM 1841[1]) were constructed in modified polylinkers of pUC18/19[2]. The DNA sequence of pRT100 between its two HindIII sites is shown; capital letters indicate CaMV sequences. TATA-box, ATG, and polyadenylation signal (underlined) as well as the putative transcriptional start (⌐) and stop (¬) are marked (ref.[3]). Transcriptional fusions are possible with pRT101 containing ACctcgagaattcgaggtc, whereas pRT100, 102, 103, 104 lead to high expression of proteins initiated at the respective ATG codon (NcoI site). In pRT103 and pRT104 (ccgaattcgg inserted into SmaI of pRT103) the ATG codon is embedded in the consensus sequence for optimal ribosome initiation in eukaryotes[4] (ACctcgagtgg*ccaccatgg*).





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[2] Yanisch-Perron, C. et al. (1985) Gene 33, 103-119
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[4] Kozak, M. (1984) Nucl. Acids Res. 2, 857-872

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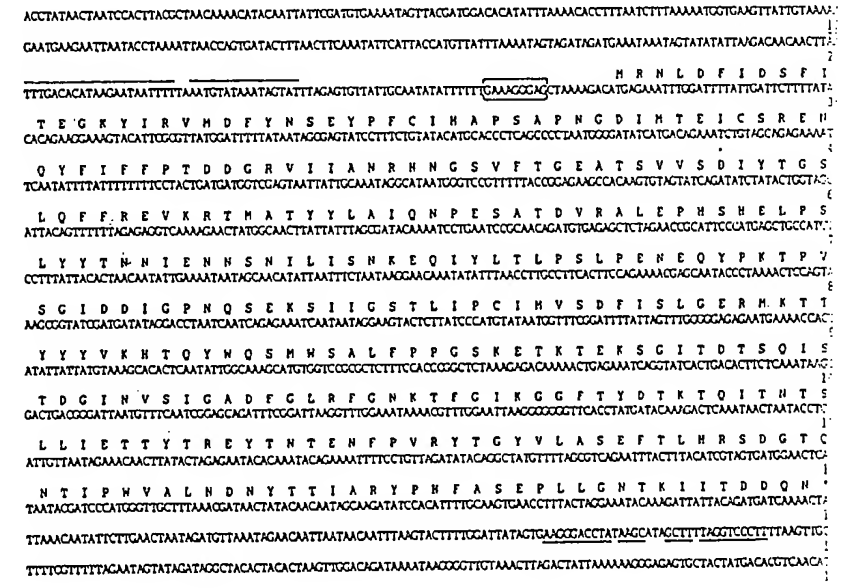
Oligonucleotide probes designed on the basis of the known N-terminal 40 a-acid sequence of the B.sphaericus 2362 larvicidal toxin (1) were used to identify an EcoRI-HindIII fragment containing the entire coding sequence. Sequence analysis showed an ORF of 1110 nucleotides corresponding to a 41 kDa protein, in agreement with an estimated 43-kDa by gel electrophoresis (1). Features of note are (i) an additional four amino-acids at the N-terminus which are not found in the purified toxin, (ii) homology of the tetrapeptide with that deduced for the N-terminus of B.thuringiensis var. israelensis and morrisoni (2), i.e. MRNL and MENL respectively, (iii) Cys replacing a reported Ser at residue 31 (no.27 in ref.1), (iv) extensive homology in the upstream control regions to B.t. subsp. kurstaki and israelensis (overlined). The putative Shine-Dalgarno sequence is boxed a a downstream inverted repeat underlined.



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(2) Earp, D.J. and Ellar, D.J. (1987). Nucleic Acids Res. 15: 3619.